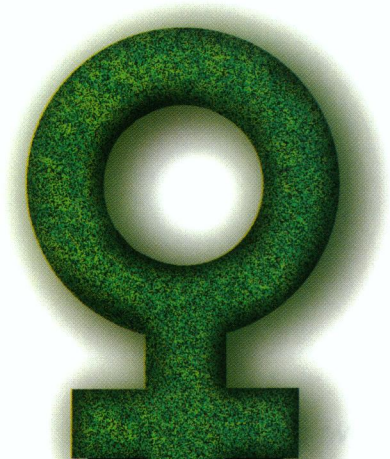
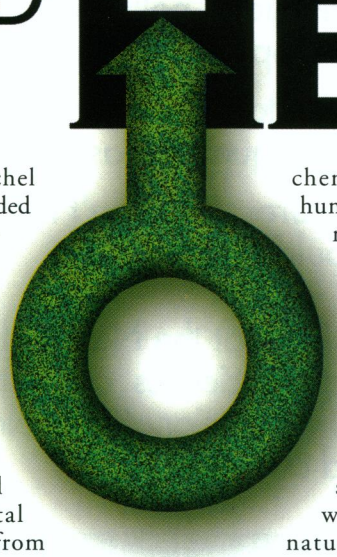


HORMONES AND HEALTH



The landmark publication of Rachel Carson's *Silent Spring* in 1967 sounded the alarm for harmful effects of chemicals on wildlife. The 1996 publication of *Our Stolen Future* by Theo Colborn, Dianne Dumanoski, and John Peterson Myers was a clarion call for recognition of similar effects on humans.The authors of *Our Stolen Future* revealed how many of the findings in animals correlate with disturbances in normal reproductive and developmental processes in humans, citing data from decades of research on wildlife that traced birth defects, sexual abnormalities, and reproductive failures to synthetic chemicals that mimic natural hormones. There is increasing evidence that environmental hormones may be linked to increases in hormone-related cancers, endometriosis, certain behavioral aberrations, and reproductive effects such as an alarming apparent drop in male sperm counts worldwide in recent years.

Recent advances in technologies have extended our ability to detect minuscule amounts of chemicals in the body and the environment, and have enabled scientists to decipher biochemical processes at the molecular level. These advances are opening new doors to understanding the complex roles endogenous and exogenous chemicals play in human health. New molecular tools are enabling researchers to study the basic mechanisms of reproductive hormones, how damaged or mutated genetic systems interact with synthetic and naturally occurring hormones, and the role such hormones play in initiating tumorigenesis, causing developmental abnormalities, and impairing reproduction. Knowledge gained from this basic research will provide critical information for developing new clinical applications, therapies, and prevention strategies.

The Endocrine System

Along with the nervous system, the endocrine system, which is composed of glands that secrete chemical messengers, is one of two communication systems that regulate all responses and functions of the body. Unlike the nervous system, which sends rapid-fire signals via electrochemical transmission along neuron conduits to the brain where they are deciphered and relayed to appropriate parts of the body to elicit instantaneous responses, the endocrine system provides cellular instructions more subtly and slowly via chemical messengers. Hormones that are produced in the endocrine glands in one part of the body travel through the bloodstream until they encounter special receptors with which they interact to initiate essential biological responses in specific target tissues. Hormones are slow-acting and their effects tend to persist in the body for long periods of time. Hormones are also specific. Each hormone has a unique chemical conformation that matches a particular receptor protein at its target cell. When a particular hormone comes in contact with its specific receptor, the two fit together as precisely as a lock and key. However, flexibility is also a feature of hormones. A particular receptor protein can be present on different kinds of cells in different organs, which means the same hormone can be used by the body to achieve different effects in different tissues. A striking feature of hormones is that dramatic changes in cellular activity are caused by extremely small amounts of chemical.

The major endocrine glands of the body include the pituitary, which regulates the other endocrine glands; the thyroid, which regulates metabolic rate; the parathyroids, which regulate blood calci-

um; the adrenals, which regulate the body's fluid and sodium balance and act as an emergency warning system in times of stress; the pancreas, which regulates blood sugar; the pineal gland, which is believed to regulate biorhythms and moods and stimulate the onset of puberty; and the gonads (ovaries in females and testes in males), which control the development of secondary sex characteristics and functioning of sex organs. As research continues, scientists are discovering that other organs and tissues, including the heart and bones, contain cells that secrete hormones essential to normal physiological function.

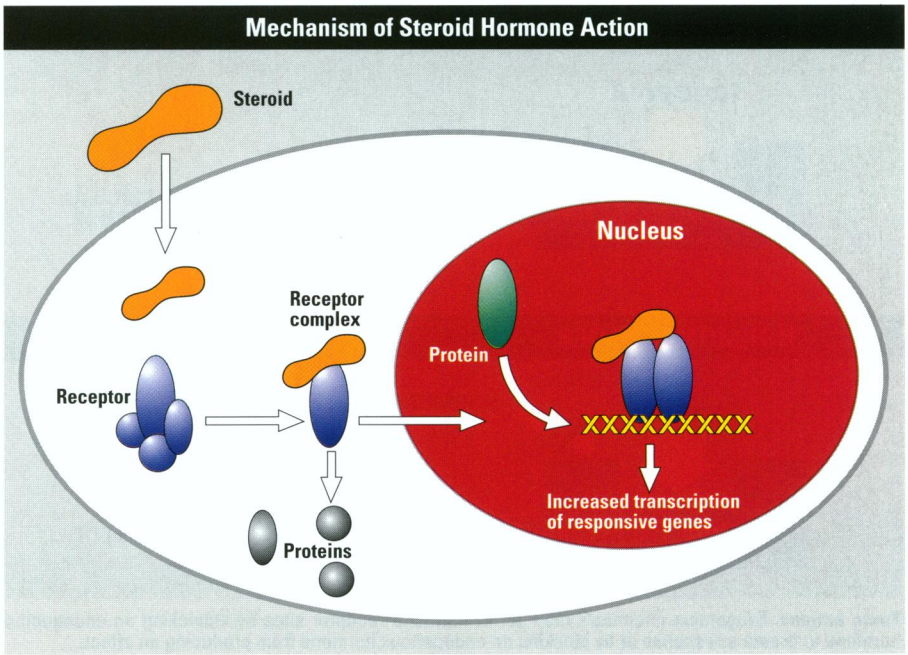
Androgens and Estrogens

Androgens and estrogens are steroids that are fat-soluble and can readily pass through the plasma membranes of their target cells. Once inside the cell, steroid hormones bind to a receptor protein and form a receptor–hormone complex that then enters the cell nucleus and binds to the DNA. The DNA binding causes a change in the gene's expression and thus changes in the cell's activity. Certain steroid hormones stimulate genes to synthesize specific proteins; others block or repress protein synthesis. There is an array of hormonally activated events—including mutations in DNA nucleotides, alteration of the chemical structure of hormones or their receptors, competition for or blocking of receptor binding sites by endogenous or environmental chemicals that mimic hormones, and a natural decline in hormone levels as a result of aging—that can initiate cellular changes, leading to cancer and irregularities of the reproductive system.

Androgens include the male hormones testosterone and androsterone and promote development of male sex characteristics. They also contribute to generalized anabolic functions of bone growth and increases in protein synthesis, especially in muscles. Androgens, like all steroid hormones, are derived from cholesterol. The principal androgen of the testes is testosterone. The production of testosterone is influenced by gonadotropic hormone dispatched from the pituitary gland (tropic hormones stimulate their target organs to secrete other hormones), and is regulated by interstitial–cell stimulating hormone (ICSH). Negative feedback from certain concentrations of testosterone in the blood lowers or blocks ICSH production. In adulthood, testosterone is essential for sperm production.

In the female, the ovaries produce two groups of steroid hormones, estrogens and progesterone. Estrogens, including estradiol, estrone, and estriol, are extremely important in the development of secondary sex characteristics and regulation of the menstrual cycle. Estrogens also influence libido and the metabolism of electrolytes and nitrogen, and help maintain pregnancy and prepare the breasts for lactation. Progesterone, which resembles estrogen chemically, helps regulate changes that occur during menstruation and influences the development of fetal membranes and mammary glands during pregnancy. Three gonadotropic hormones produced by the pituitary gland regulate the secretion of estrogens and progesterone: follicle-stimulating hormone (FSH), luteinizing hormone (LH), and prolactin.

Some male and female hormones are actually secreted by both sexes. Male hormones are secreted in greater quantities and are more potent when produced by the male, and the same is true for female hormones when produced by females.



Protein producers. In a general model, steroid hormones passively enter a cell and bind to a receptor to form a receptor complex, releasing proteins in the process. Other proteins may also bind to the receptor complex which permits interactions with specific DNA sites to cause transcription of responsive genes.

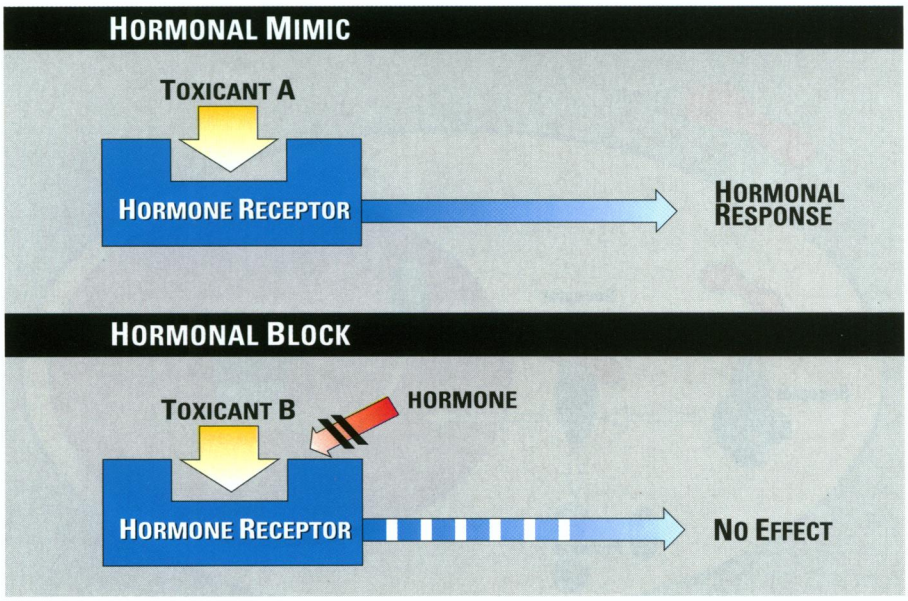
Hormone Function and Regulation

Hormone production is regulated by a complicated negative-feedback pathway that is turned on and off in response to fluctuating hormone levels. When hormone production peaks, the hormone acts as an inhibitor and causes the pathway producing the substance to shut down. At the top of the chain of command is the hypothalamus. This part of the brain coordinates signals between the central nervous and endocrine systems by releasing factors that initiate a chemical cascade. The cascade switches on subsequent hormone production in target cells of other endocrine glands or tissues in the body.

Once hormones are produced, they travel through the bloodstream to target cells where they initiate a change in cellular activity by attaching to the receptor protein of certain enzymes. This change is transmitted across the plasma membrane of a cell in one of three ways, depending on the type of hormone. The molecule may bind to a receptor protein that carries it across the membrane into the cytoplasm of the cell. A molecule may move directly into the nucleus, where it binds to its receptor protein and initiates transcription of messenger RNA. In the case of peptide hormones, which cannot pass through the plasma membrane, the molecule binds to receptor proteins on the surface of the cell. Protein receptor binding triggers a change in the shape of the receptor protein and unleashes a series of events in the cytoplasm. Activation or deactivation of chemical messages occurs as the signal is passed from one molecular receptor to the next.

Hormonal Carcinogenesis

Sex hormones and other steroids alter gene expression in target cells. In the absence of a steroid, an inhibitor protein prevents the hormone receptor from binding to the DNA and inhibits transcription of mRNA. But when a steroid binds with its receptor, it releases the inhibitor protein and the receptor can bind to regulatory sites on the DNA, thereby affecting expression of certain genes and their protein products in



Toxic actions. Exogenous chemicals may act at hormone receptor sites by mimicking an endogenous hormone to create a response or by blocking an endogenous hormone from producing an effect.
Source: McLachlan JA. Functional toxicology: a new approach to detect biologically active xenobiotics. Environ Health Perspect 101:386–387 (1993).

the target cell. Because sex hormones play important roles in regulating the growth and differentiation of cells in the sex organs, changes involving hormones, their receptors, and transcription factors that interact with estrogen and androgen receptors may lead to hormonal carcinogenesis.

A hormonal role in carcinogenesis was first reported by G. T. Beatson in an 1898 *Lancet* article, which described a reduction of breast cancer in women whose ovaries had been surgically removed, resulting in greatly lowered estrogen levels. Subsequent studies in rodent models and other systems have repeatedly shown that estrogen induces tumor formation in various organs and tissues including the breast, uterus, prostate, and kidney. Although it is clear hormones are involved in increased cell proliferation and hormone-initiated carcinogenesis, the biochemical and molecular mechanisms involved in the initiation and promotion of tumorigenesis are not fully understood. Some researchers believe hormones are genotoxic and directly involved in altering DNA structure; others propose a more indirect role via oxidation of reactive metabolites formed during sex hormone metabolism.

Androgens. There is debate about whether hormonally induced cancers result from direct alteration of genetic material by hormones (or products of hormone metabolism) or from an indirect influence of hormones on DNA via cellular activities. In order to elucidate the molecular mechanisms of hormonal carcinogenesis, Jonathan J. Li, director of the Division of Etiology and Prevention of Hormonal Cancers at the Kansas Cancer Institute, and colleagues have been investigating estrogen-induced kidney tumors in rodent models. Li's group has shown that exposure to natural and synthetic estrogens has a cytotoxic effect on cells, resulting in damage to kidney tubules. Genetic instability, as evidenced by chromosomal breaks, DNA adducts, and inappropriate expression of growth-promoting proto-oncogene products and tumor suppressor gene products after exposure to estrogen, is linked to cell proliferation in response to injury. These findings point to a hormonally mediated epigenetic role for sex steroids rather than a metabolically activated genotoxic role in gene regulation. Says Li, "In recent years, the data on metabolism of estrogen indicate it doesn't play much of a role in carcinogenesis."

Some researchers believe that oxygen free radicals—electrically charged molecules in which the number of electrons is not equal to the number of protons—generated by estrogen metabolism may play a major role in damaging reproductive tract tissue. Such molecules are unstable and highly reactive because they must balance their charges; this reactivity leads to cellular damage and cell death. Over years of exposure to free radicals, cumulative damage to reproductive tract tissue could enhance susceptibility to the onset and development of cancerous lesions.

A clear correlation between sex hormones and prostate cancer linked to genotoxic activity of estrogens in combination with androgen was reported by Maarten C. Boslund, associate professor of environmental medicine and urology at the New York University Medical Center in Tuxedo, New York, in the 1996 book *Cellular and Molecular Mechanisms of Hormonal Carcinogenesis: Environmental Influences.* As reported by Boslund, administration of testosterone to young, castrated, male rats caused 20–30% development of prostate tumors, whereas treatment with testosterone and estrogen combined produced prostate cancer in 100% of the treated rats. These tumors formed in regions where there was increased oxidative tissue damage, signaling a connection between oxidative damage and carcinogenesis.

Shuk-mei Ho, an associate professor of biology at Tufts University, is using a rodent model system with features of the human prostate to study how free radicals produced by estrogen metabolism can cause DNA damage and initiate tumorigenesis. Long-term treatment of Noble rats (a strain highly susceptible to hormone-induced carcinogenesis) combining androgen with estrogen produced an increase over time in cell proliferation that significantly exceeded that of rats treated with androgen alone. In humans, as males age, androgen production declines but estrogen levels remain the same or in some cases

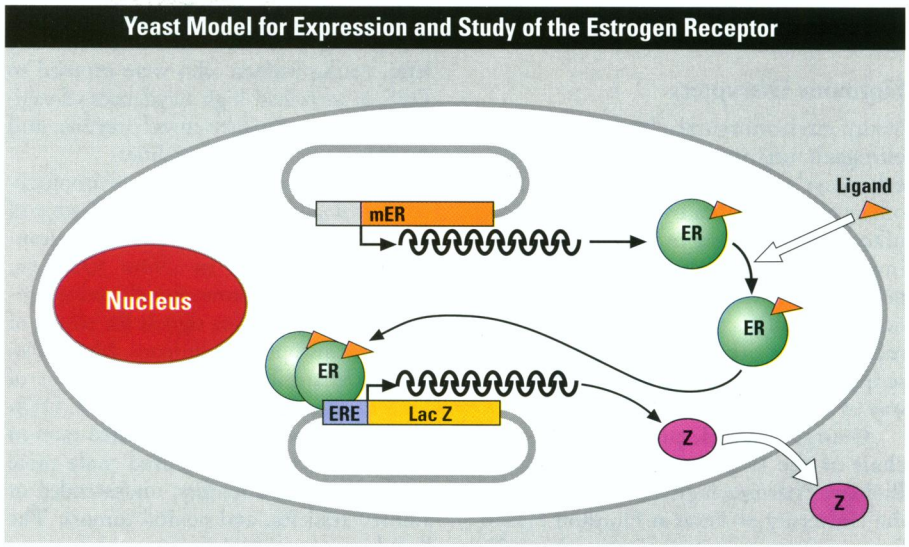

Shuk-Mei Ho

A Noble profession. Scientists are using Noble rats—a strain highly susceptible to hormone-induced carcinogenesis—to study the role of free radicals in initiating prostate tumors.

may increase. Increases in prostate cancer in older men may be related to this modulation of sex hormone levels and concomitant increases in cell damage by metabolites from estrogen oxidation. Ho's research, published most recently in the 1997 book *Prostate: Basic and Clinical Aspects*, shows that enzymes that modulate tissue destruction by free radicals also have impaired function in prostate cancer cells, indicating that there may be a synergistic relationship between DNA mutation and metabolic conversion products that causes cell damage and a proliferation of prostate carcinogenesis. According to Ho, "Metabolic conversion can activate a sex hormone to become a chemical carcinogen that interacts with and can alter DNA. Anything in the environment that can mimic this process will elevate oxidative stress and cause cancer."

The complexity of how hormones are related to carcinogenesis is highlighted by research showing that mutations in the androgen receptor change the steroid or DNA-binding activities of that receptor, which can lead to prostate cancer completely independent of the hormone. Dolores J. Lamb, associate professor of urology and cell biology at the Baylor College of Medicine, and colleagues have shown that a mutated androgen receptor (AR) gene can be activated by an enzyme in the absence of androgen, indicating that alternative signaling pathways may activate tumorigenesis.

The importance of the AR gene for normal sexual development was revealed by a study of five subjects with ambiguous reproductive organs. The study was published by reproductive biologist Terry Brown and colleagues from the Division of Reproductive Biology at Johns Hopkins University in a 1996 research article in *Molecular and Cellular Endocrinology*. Genetic analysis by Brown found missense mutations in the AR genes that prevent androgen-receptor binding to DNA. Consequently, all five subjects were insensitive to androgen and did not undergo normal male sexual determination and differentiation. Brown's lab has pinpointed natural mutations in two regions of the AR gene that encode amino acid substitutions in the DNA-binding domain that are linked to complete androgen insensitivity. This work underscores the importance of the AR gene for normal sexual development. Therefore, any chemical that induces mutation in the AR gene or acts as an analogue that binds and represses AR activity may have serious deleterious effects on embryogenesis.



Tell-tale signs. In an estrogen expression model, estrogen response elements (EREs) enable mouse estrogen receptor (mER) to bind to target genes in yeast, producing reporter protein (Z) that can be measured. Such models are being used to test the interaction of the ER and toxic chemicals.

Estrogens. Estrogen induces the growth of epithelial tissue in the uterus and mammary glands and has been implicated in breast cancer. Anatomist Gerald Cunha of the University of California at San Francisco is investigating the cellular and molecular mechanisms of steroid sex hormones in the epithelial tissue that lines reproductive organs. Using genetically engineered knockout mice that lack the estrogen receptor (ER) gene, Cunha has demonstrated that mammary gland growth is mediated by binding to an ER in the stroma, or connective tissue of the breast. Additional experiments using knockout mice for the ER growth factor indicate there is a complex molecular pathway of interactions in which estrogen acts on mammary stromal cells via the ER to elicit a crucial estrogen growth factor receptor that in turn elicits the release of another factor causing growth of the mammary epithelial cells. New evidence is showing how multiple steps in the pathway may be affected by different chemical compounds including endogenous proteins and environmental toxins, and how injury during these steps may have various effects on human health.

Jan-Åke Gustafsson, a medical nutritionist at the Karolinska Institute in Huddinge, Sweden, is studying how estrogen receptors function to modulate the growth and development of the ovaries. In the June 1996 issue of the *Proceedings of the National Academy of Sciences*, Gustafsson characterized two estrogen receptors, ERα and ERβ. Binding to the ER is critical to the expression of genes that contribute to normal ovarian function. To determine if both ER genes are expressed equally in the ovary, experiments, reported in the February 1997 issue of *Molecular Endocrinology*, were conducted using transgenic rats with an ERα knockout gene. Because ovary follicle growth was present in rats lacking ERα, it appears that the ovary expresses estrogen-binding molecules other than ERα, indicating that ERβ may play a role in estrogen binding in the ovary. Comparison of expression of the two types of ERs in the ovary and uterus showed that higher levels of ERβ are expressed in the uterus and higher levels of ERα are expressed in the ovaries. Furthermore, expression of ERs is modulated by the ability of estrogen to elicit responses in other target genes such as the progesterone receptor (PR), growth factors, and gonadotropic hormones. The differences in expression and relative ligating ability of the different forms may explain the selectivity of ER agonists and antagonists, such as follicle-stimulating hormone (FSH), luteinizing hormone (LH), or prolactin, in different target tissues.

An interesting and unexpected development of Gustafsson's work in locating "orphan" receptors—receptors for which no known ligand has been identified—was the discovery of an ER in the prostate. The group was searching for novel nuclear receptor proteins and found a new receptor that had a high affinity for binding estrogen. The group has since identified the human homologue of the ER found in the rat prostate. Whereas estrogen was once thought to function mainly in reproduction, discoveries of new estrogen receptors are signaling important roles for estrogen related to other aspects of human health,

such as bone maintenance and the cardiovascular system.

Hormone Disrupters

Many environmental chemicals exhibit estrogenic activity. Some of these chemicals occur naturally in plants and fungi, others are man-made by-products of manufacturing, present in agricultural and industrial chemicals. Estrogenic compounds, both environmental and endogenous, may interact with estrogen, estrogen receptors, or other hormones and transcription factors in the biochemical pathway of hormone activity.

George Stancel, professor and vice chair of the Department of Integrative Biology, Pharmacology, and Physiology at the University of Texas at Houston Health Science Center, is investigating how the hormone-receptor complex affects transcription through binding with nucleotide sequences known as estrogen receptor elements (EREs). EREs are sequences of 12 nucleotides that enable the receptor to bind to target genes and are found in the regulatory regions of hormone-responsive genes. The finding that there are distinct EREs for two genes that are members of a family of transcription factors (reported in the October 1995 supplement to *EHP*) led Stancel to hypothesize that there are different interactions of the ER with DNA sequences that can initiate transcription of various nuclear genes in a cell. This could create different dose–response curves for various biological responses due to the regulation of multiple genes by estrogens. For example, environmental estrogens could bind to an ER and either block or enhance transcription; they could cause toxicity by directly altering DNA structure; or they might be involved in modulating an imbalance in levels of hormonally regulated growth factors, growth factor receptors, proto-oncogenes, or other target molecules in the molecular interactions involved in growth and differentiation of hormone-responsive cells.

Numerous examples of reproductive abnormalities in wildlife caused by environmental contaminants that have estrogenic properties have been described. In one instance, alligators exposed to runoff of pesticides in Lake Apopka, Florida, exhibited a range of reproductive abnormalities including reduced fertility, smaller-than-normal penises, and shifts in sex ratios with predominantly females being born.

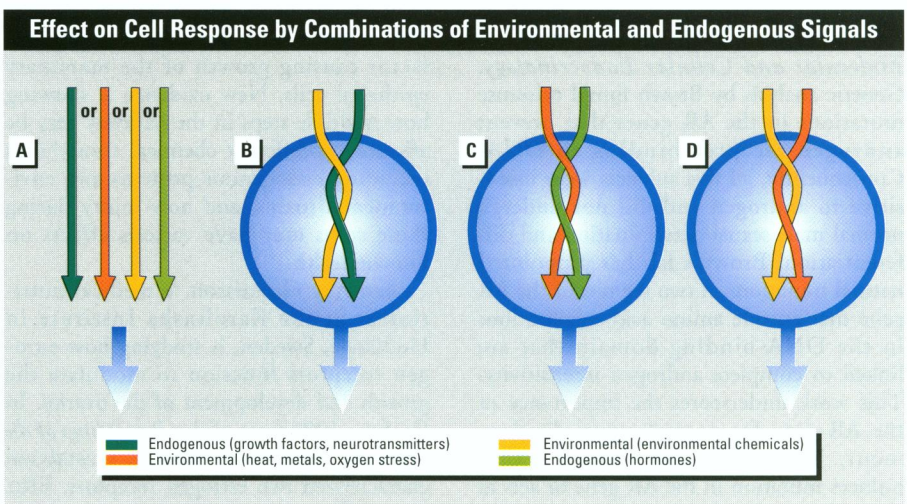
Diethylstilbestrol (DES), a synthetic chemical that acts like natural estrogen and that was widely prescribed to pregnant women in the 1950s and 1960s to prevent miscarriage, among other uses, provides an early example of the effects of hormone-disrupting chemicals on humans. Years later, young women who were exposed to DES *in utero* had high incidences of vaginal cancer and experienced sterility and reproductive tract abnormalities.

Research in the 1970s by environmental toxicologist John McLachlan, director of the Tulane/Xavier Center for Bioenvironmental Research in New Orleans, Louisiana, showed how animal studies provide clinically relevant signals for effects of hormone disruptors in humans. McLachlan replicated similar reproductive effects of DES in experimental mouse models. DES-exposed female mice had increased rates of vaginal cancer, and exposed male mice exhibited reduced fertility, undescended or stunted testicles, and genital tumors. The link between animal studies and humans gained significance years later when large numbers of adults who had prenatal exposure to DES had difficulties having children and developed cancers. More recently, McLachlan has investigated the possibility of synergistic interactions between environmental estrogens. Although xenobiotic hormones normally have a much lower binding affinity than the body's own natural estrogen, exposure to combinations of some chemical mixtures may cause a dramatic rise in estrogenic activity. In experiments by McLachlan and colleagues, reported in the 7 June 1996 issue of *Science,* a combination of the weakly estrogenic pesticides endosulfan, dieldrin, toxaphene, and chlordane, measured by yeast cell ER binding assays, produced estrogenic activity 150–1,600 times greater than for any of the chemicals alone. Synergistic responses were also observed in mammalian cell culture experiments. According to McLachlan, such magnification of effect signals the potential for an extreme health hazard to humans who are exposed to combinations of specific environmental chemicals. In addition to greatly magnified quantitative effects, synergism can amplify qualitative effects when different chemicals act on different signaling pathways involved in the hormone action in target cells.

In a technical comment on McLachlan's study, published in the 17 January 1997 issue of *Science*, Kavita Ramamoorthy and colleagues in the Texas A&M University Department of Veterinary Physiology and Pharmacology reported their failure to replicate synergistic interactions between dieldrin and toxaphene in ER-binding experiments in immature female mice, human breast cancer cell lines, and yeast assays. The group's results indicated that estrogenic effects are additive rather than synergistic, and therefore mixtures of two compounds can be expected to be weakly estrogenic and will not have a dramatic adverse environmental and human health impact. Because of the concern over harmful effects of environmental chemicals, the conflicting results of the two laboratories' studies have sparked a heated debate about synergistic effects of organochlorine pesticides. However, as McLachlan and colleagues point out in a reply to Ramamoorthy in the same issue of *Science*, there are important differences in the design of the experiments conducted in the two labs that may ultimately shed new light on the complexity of the molecular biology of hormone interactions.

Clinical Implications

As data emerge from many avenues, concerns about the risks to human health



The interaction of a cell with an endogenous or environmental signal (A) produces a response. The interaction of a cell with a combination of environmental and endogenous signals (B,C, or D) may result in a synergistic response.

Source: Arnold SF, McLachlan JA. Synergistic signals in the environment. Environ Health Perspect 104:1020–1022 (1996).

posed by endocrine disruptors have led to the identification of many such environmental chemicals. Objectives of ongoing research are to continue to identify potentially harmful chemicals, define molecular and physiological mechanisms of action, clarify mutagenic properties and genetic susceptibility, specify risk factors from exposure, and develop therapies and prevention strategies.

Assay for toxicity screening. McLachlan's group is developing ways to characterize estrogenic properties of environmental chemicals that would parallel the diagnostic capabilities of the Ames test used in screening chemicals for carcinogenicity. The group has developed three types of *in vitro* tests that have yielded promising results. A yeast estrogen screen (described in the May 1996 issue of *EHP*) was developed to measure enzyme levels from transcription of the human ER (hER) and two estrogen response elements linked to a reporter gene in yeast. Direct interaction of a chemical compound with hER is tested first in a competition binding assay. Next, estrogenic activity is measured in an estrogen-sensitive, transgenic human breast cancer cell line that carries two EREs linked to the luciferase reporter gene. Using this method to screen for the known xenoestrogens DES and DDT showed enhanced enzyme activity. McLachlan advocates the use of *in vitro* assays in conjunction with whole-animal models to evaluate environmental chemicals for hormone-disrupting capabilities.

Biomarkers of exposure. Oncologist Saraswati Sukumar of the Johns Hopkins Oncology Center is working on finding a genetic marker for the early detection of breast cancer. The ends of chromosomes, called telomeres, carry a specific set of DNA nucleotides that ensure the integrity of the chromosome. If the ends of the chromosome are broken or damaged, these nucleotides can become "sticky" and fuse with other chromosomes, causing misalignments and rearrangements of genetic material. In response to such damage, the cell produces lots of telomerase, the enzyme involved in repairing telomeres. Because this process occurs in cancer cells and not in normal human cells, Sukumar's group is measuring levels of telomerase activity in the fluid of cancer cells as a potential biomarker for breast cancer by investigating induction of telomerase activity in rat models following exposure to the carcinogenic compound N-methyl-N-nitrosourea.

Work on tumor-suppressor genes by Carrie W. Rinker-Schaeffer, director of urologic research and associate director of the Prostate Cancer Program at the University of Chicago Cancer Research Center, may lead to biomarkers for detecting the onset of prostate cancer. Identification of the *KAI1* gene on chromosome 11, which suppresses prostate tumor metastasis, has enabled Rinker-Schaeffer to study expression of this gene in comparative assays using normal human cells with prostate cancer cell lines. *KAI1* RNA is strongly expressed in normal cells, but down-regulated in cancer cells, so it may provide a molecular tool to screen for prostate cancer. According to Rinker-Schaeffer, "Every three minutes an American man is diagnosed with prostate cancer, and every hour four men will die of this disease. The enormity of this problem demonstrates the critical need for markers that will allow for accurate diagnosis and treatment."

Treatment strategies. It is well established that estrogen enhances cell growth and proliferation in breast cancer. A first-line chemotherapy in the treatment of hormone-dependent breast cancer is to block estrogen activity with the drug tamoxifen. Although initially effective, patients develop a resistance and disease progression resumes. Angela Brodie, professor of pharmacology and experimental therapeutics at the University of Maryland School of Medicine, is developing new therapies based on mechanisms of action for hormones and hormone receptors. Brodie is interested in two pharmacologic approaches to reducing estrogen effects: inhibition of estrogen activity with antiestrogens that bind ERs in the tumor, and inhibition of estrogen production using chemicals that block the production of aromatase, an enzyme that synthesizes estrogen. A variety of aromatase inhibitors undergoing clinical testing promise to lead to new drugs for the treatment of breast cancer.

Prevention strategies. Phytoestrogens, naturally occurring estrogens found in plants, have been used for thousands of years by indigenous medical practitioners. In contrast to xenoestrogens from industrial pollutants that bioaccumulate in animal and human tissues and may cause adverse health effects, phytoestrogens have a long history of coevolution with humans and animals and are quickly metabolized by endogenous enzymes. The low incidence of hormone-related cancers, osteoporosis, heart disease, and menopausal symptoms in certain populations with diets rich in phytoestrogens has led to renewed interest in the potential beneficial effects of phytoestrogens in treating and preventing disease. Asian populations consume a diet rich in soy products, which contain the phytoestrogens genistein and daidzein. Because of evidence of lesser incidence among such populations of hormonally related cancers, and because there is a greater incidence of hormonally related cancers among Asians who have migrated to the United States and consequently changed their dietary habits, it seems possible that phytoestrogens in the diet may be useful in the prevention of certain estrogen-related diseases.

Evidence to support this thesis was provided by Coral Lamartiniere, professor of pharmacology and toxicology and director of the graduate training program in toxicology at the University of Alabama, who showed that genistein, an isoflavone phytoestrogen, protects against chemically induced mammary cancer in an animal model. Neonatal and prepuberty rats were exposed to genistein by injection. In early adulthood, treated rats were dosed with a high concentration of the carcinogen dimethylbenz[*a*]anthracene (DMBA). A 50% reduction in development of tumors was observed in genistein-exposed rats, and there was a longer latency period for tumor development. Results from subsequent experiments soon to be published reveal that protection against DMBA-induced mammary cancer by phytoestrogens is dose-dependent. In contrast with deleterious effects on reproductive development from neonatal exposure to xenohormones, early exposure to phytoestrogens may confer protection against breast cancer later in life. As Lamartiniere points out, "For the longest time, scientists have viewed exposures to external chemicals during the prenatal period as potential for toxicity, but this opens the opportunity for beneficial 'programming' against future exposures to environmental toxins."

New technologies and approaches, including enhanced tools for understanding molecular interactions and mechanisms, are helping scientists to understand the often powerful effects—both adverse and beneficial—that very small amounts of environmental hormones and hormone-like chemicals may have on humans and wildlife. Scientists exploring the endocrine system and the ramifications of its interactions with environmental chemicals will ultimately provide the basis for determining risk factors from exposure, promote debate, and generate public policy in regard to the use and distribution of chemicals in the environment.

Mary W. Eubanks